



HAL
open science

Genomic study of maternal lipid traits in early pregnancy concurs with four known adult lipid loci

Marion Ouidir, Suvo Chatterjee, Jing Wu, Fasil Tekola-Ayele

► To cite this version:

Marion Ouidir, Suvo Chatterjee, Jing Wu, Fasil Tekola-Ayele. Genomic study of maternal lipid traits in early pregnancy concurs with four known adult lipid loci. *Journal of clinical lipidology*, 2024, 17 (1), pp.168 - 180. 10.1016/j.jacl.2022.10.013 . hal-04662335

HAL Id: hal-04662335

<https://hal.science/hal-04662335v1>

Submitted on 25 Jul 2024

HAL is a multi-disciplinary open access archive for the deposit and dissemination of scientific research documents, whether they are published or not. The documents may come from teaching and research institutions in France or abroad, or from public or private research centers.

L'archive ouverte pluridisciplinaire **HAL**, est destinée au dépôt et à la diffusion de documents scientifiques de niveau recherche, publiés ou non, émanant des établissements d'enseignement et de recherche français ou étrangers, des laboratoires publics ou privés.



Distributed under a Creative Commons Attribution - NonCommercial - NoDerivatives 4.0 International License



Published in final edited form as:

J Clin Lipidol. 2023 ; 17(1): 168–180. doi:10.1016/j.jacl.2022.10.013.

Genomic study of maternal lipid traits in early pregnancy concurs with four known adult lipid loci

Marion Ouidir, PhD¹, Suvo Chatterjee, PhD¹, Jing Wu, PhD², Fasil Tekola-Ayele, PhD¹

¹Epidemiology Branch, Division of Population Health Research, Division of Intramural Research, Eunice Kennedy Shriver National Institute of Child Health and Human Development, National Institutes of Health, Bethesda, MD, USA

²Division of Population Health Research, Division of Intramural Research, Eunice Kennedy Shriver National Institute of Child Health and Human Development, National Institutes of Health, Bethesda, MD, USA

Abstract

Background: Blood lipids during pregnancy are associated with cardiovascular diseases and adverse pregnancy outcomes. Genome-wide association studies (GWAS) in predominantly male European ancestry populations have identified genetic loci associated with blood lipid levels. However, the genetic architecture of blood lipids in pregnant women remains poorly understood.

Objective: Our goal was to identify genetic loci associated with blood lipid levels among pregnant women from diverse ancestry groups and to evaluate whether previously known lipid loci in predominantly European adults are transferable to pregnant women.

Methods: The *trans*-ancestry GWAS were conducted on serum levels of total cholesterol, high-density lipoprotein cholesterol (HDL), low-density lipoprotein cholesterol (LDL) and triglycerides during first trimester among pregnant women from four population groups (608 European-, 623 African-, 552 Hispanic- and 235 East Asian-Americans) recruited in the NICHD Fetal Growth Studies cohort. The four GWAS summary statistics were combined using *trans*-ancestry meta-analysis approaches that account for genetic heterogeneity among populations.

Results: Loci in *CELSR2* and *APOE* were genome-wide significantly associated (p-value < 5×10^{-8}) with total cholesterol and LDL levels. Loci near *CETP* and *ABCA1* approached genome-wide significant association with HDL (p-value = 2.97×10^{-7} and 9.71×10^{-8} , respectively). Less than 20% of previously known adult lipid loci were transferable to pregnant women.

Correspondence: Fasil Tekola-Ayele, PhD, Epidemiology Branch, Division of Population Health Research, Division of Intramural Research, Eunice Kennedy Shriver National Institute of Child Health and Human Development, National Institutes of Health, 6710B Rockledge Drive, Room 3204, Bethesda, MD 20892-7004, ayeleft@mail.nih.gov.

Author Contributions Statement: FT-A conceived the research idea and contributed to the interpretation. JW conducted quality control of the genome-wide data. MO conducted data analysis, drafted and revised the manuscript. SC, JW and FT-A provided critical intellectual content. All authors approved the final version of the manuscript.

Publisher's Disclaimer: This is a PDF file of an unedited manuscript that has been accepted for publication. As a service to our customers we are providing this early version of the manuscript. The manuscript will undergo copyediting, typesetting, and review of the resulting proof before it is published in its final form. Please note that during the production process errors may be discovered which could affect the content, and all legal disclaimers that apply to the journal pertain.

Disclosures: none.

Conclusion: This *trans*-ancestry GWAS meta-analysis in pregnant women identified associations that concur with four known adult lipid loci. Limited replication of known lipid-loci from predominantly European study populations to pregnant women underlines the need for genomic studies of lipids in ancestrally diverse pregnant women.

Clinical Trial Registration: [ClinicalTrials.gov](https://clinicaltrials.gov/ct2/show/study/NCT00912132), NCT00912132.

Keywords

Pregnancy; Lipids; Genomics; Cholesterol; LDL; GWAS; Multi-ancestry

INTRODUCTION

Women's blood lipid levels undergo physiological changes during pregnancy compared to pre-pregnancy levels, and the changes are apparent in early gestational weeks (1, 2). While these lipid metabolic changes during pregnancy are crucial for fetal development and maintenance of pregnancy, abnormal blood lipid levels during pregnancy are associated with increased risk for adverse pregnancy outcomes (3, 4), aberrant fetal growth (5–9), and later-life cardiovascular diseases for the mother (10), and dyslipidemia and cardiovascular diseases in the offspring during childhood and adulthood (11–13). Epidemiological and family studies suggest that physiological lipid changes during pregnancy are determined by both environmental and genetic factors (14–16). Therefore, understanding the biological mechanisms that underlie genetic regulation of lipid metabolism during pregnancy can shed light on a broad array of pregnancy complications and health outcomes across the lifespan. Such studies, however, are scarce.

The heritability of blood lipid traits in adults involving men and non-pregnant women has been estimated to be 30–80% (17–22). Genome-wide association studies (GWAS) conducted in cohorts with predominantly male European ancestry participants have identified genetic loci related to blood lipid traits (23–33). Including diverse ancestral populations in GWAS of lipid traits is useful to identify ancestry-shared as well as ancestry-specific genetic associations (24, 28, 29), address limited cross-ancestry transferability of lipid trait loci (34), and improve the accuracy that lipid trait polygenic scores to predict risks of cardiovascular diseases and fetal growth outcomes during pregnancy (9, 35, 36). Accumulating evidence shows that lipid metabolism and blood lipid traits in women differ from men, although the underlying mechanisms are not clearly understood (37, 38). Moreover, studies have shown that some lipid trait-related loci show effect differences by sex (18, 31, 39) and the heritability of lipid traits varies by sex (20, 40). Most studies have reported higher heritability of HDL, LDL and triglycerides in women compared to men (18, 20, 41); however, studies have also reported lower additive heritability of LDL (18) and triglycerides (42) among females compared to males. Therefore, it is possible that the genetic architecture of blood lipid traits in pregnant women may show some differences from men, but remains poorly understood.

Our goal was to identify genetic loci associated with blood lipid traits in pregnant women from diverse ancestry populations, and to evaluate whether previously known lipid trait GWAS loci in adults are transferable to pregnant women. Furthermore, we aimed to

elucidate the mechanisms that underlie lipid trait-related genetic loci by identifying potential functional genes that share genetic signal with gene expression in relevant tissues using genetic colocalization.

MATERIALS AND METHODS

Population

Our analysis included 608 European American, 623 African American, 552 Hispanic American and 235 East Asian American pregnant women as self-identified predominant race/ethnic groups from the NICHD Fetal Growth Studies–Singletons. Briefly, the NICHD Fetal Growth Studies–Singletons was a prospective cohort study that included 2,802 pregnant women (European-, African-, Hispanic- and East Asian-American) to determine race/ethnic differences in fetal growth. Pregnant women were recruited between 8–13 gestational weeks between July 2009 and January 2013 at 12 clinic sites in the U.S. as previously reported (43). Pregnant women were excluded if they reported having major medical condition (such as cancer, autoimmune disease, diabetes, chronic hypertension, HIV or AIDS, chronic renal disease and psychiatric disorder). In addition, non-obese pregnant women (pre-pregnancy body mass index [BMI] lower than 29.9 kg/m²) were excluded if they reported past adverse pregnancy outcomes and self-reported consumption of cigarettes, illicit drugs or alcohol in the months prior to enrollment. Written informed consent, including the agreements of their biospecimen usage was obtained from all study participants.

Maternal blood lipid level measurement

Lipid measurement methods have been previously published (44, 45). Briefly, total cholesterol, high-density lipoprotein cholesterol (HDL) and triglyceride plasma concentrations were directly measured in maternal non-fasting blood samples collected between 10 gestational weeks 0 days to 13 gestational weeks 6 days of pregnancy using the Roche COBAS 6000 chemistry analyzer (Roche Diagnostics, Indianapolis, IN). The inter-assay laboratory coefficients of variation were 2.2%, 3.2% and 2.3% for total cholesterol, HDL and triglycerides, respectively. Low-density lipoprotein cholesterol (LDL) was calculated using the Friedewald formula (46).

Genotyping, imputation and quality control

DNA was extracted from stored buffy coat specimens obtained from pregnant women's blood at enrollment. Detailed DNA genotyping, quality control, and imputation procedures have been reported previously (47). Briefly, single nucleotide polymorphisms (SNPs) were genotyped using the Infinium Multiethnic Global BeadChip microarray (Illumina) that has ~1.7 million SNPs. Individuals with > 5% missing SNP genotypes, high degree of relatedness ($\hat{\pi} > 0.25$), and excess heterozygosity (> 3 standard deviation (s.d.) from the mean) were removed. SNPs with >5% missing values, minor allele frequency <0.5%, and not in Hardy-Weinberg equilibrium (p -value $< 10^{-4}$) and insertion-deletions, multi-allelic and duplicated SNPs were removed. Principal components (PCs) from multi-dimensional scaling analysis of a pruned set of uncorrelated genome-wide SNPs were generated using PLINK.

Ancestry-specific GWAS analysis

In each of the four population groups, linear regression analysis was performed to test the associations of maternal SNPs with each lipid trait individually (i.e. total cholesterol, LDL, HDL and triglycerides). The analyses were performed under an additive genetic model adjusting for maternal age and the first five PCs as implemented in MACH2QTL (48). Power estimation for East Asian Americans (smallest sample size, n=235) and for African Americans (largest sample size, n=623) are reported in Supplementary Figure S1.

Trans-ancestral meta-analyses

The four ancestry-specific GWAS summary statistics were combined using TransMeta that implements kernel regressions-based random effect model to account for genetic heterogeneity among ancestral populations (49). To account for population differentiation due to genetic structure, TransMeta uses a genetic similarity kernel structure (K matrix), where K is constructed prior to carrying out the data analysis based on the HapMap3 data with identified ancestry groups using CEU (referring to Utah residents with Northern and Western European ancestry from the CEPH collection) for European Americans, YRI (referring to Yoruba in Ibadan, Nigeria) for African Americans, MEX (referring to Mexican ancestry in Los Angeles, California, United States) for Hispanic Americans and CHD (referring to Chinese in Metropolitan Denver, Colorado, United States) for East Asian Americans. Quantile-quantile (QQ) plots of p-values and the corresponding genomic inflation factor (λ) are reported in Supplementary Figure S2. To account for genomic inflation, we calculated genomic controlled p-value adjusted for genomic inflation factor (λ). Genome-wide significance was set as p-value $< 5 \times 10^{-8}$. Significant results are presented using regional visualization (50) and annotated on genome build GRCh37 (51). Regression coefficients of trans-ancestry meta-analysis were obtained using GWAMA (Genome-Wide Association Meta-Analysis) (52).

Variance in lipids explained by genetic variants

We estimated the proportion of variance in each lipid trait explained by two sets of SNPs: 1) lead SNPs associated with each lipid trait in our analysis, and 2) previously published loci associated with lipids from 2 recent multi-ancestry GWAS papers (23, 24). De Vries *et al.* reported 314 known lipid loci (23). Klarin *et al.* included 297,626 multi-ethnic veterans and identified 141 leads variant associated with lipid (24). Together, 454 unique SNPs associated with lipids were identified by the two studies, of which 431 SNPs were available in at least one ancestry group in our data and 383 unique SNPs were available in all the 4 ancestry groups. The 431 unique SNPs (148 cholesterol-, 44 LDL-, 166 HDL- and 147 triglycerides-related loci) were used to compute variance in lipid traits explained by previously published loci. Variance explained was calculated to be the difference in adjusted coefficient of determination (adjusted r^2) between two regression models with and without the SNPs (i.e., model 1 adjusted for SNPs of interest, maternal age and the first five PCs and model 2 adjusted only for maternal age and the first five PCs). Variance explained was calculated for each lipid trait, per each ancestry group.

Evaluation of published lipid loci in pregnancy women

Among the 383 previously published SNPs available in all the 4 ancestry groups, 366 unique SNPs (consisting in 148, 44, 165 and 145 loci related to total cholesterol, LDL, HDL and triglycerides, respectively) with complete information in the published papers (direction of the association and reference allele) were evaluated. Two types of evaluations were performed: i) direct look-up of the previously published query SNPs (i.e. exact replication) in our trans-ancestry meta-analysis summary statistics. SNPs with p-value < 0.05 and with the same effect directions as the published lipids loci were considered to be significantly replicated. ii) look-up of the set of SNPs correlated with the published query SNPs (i.e., linkage disequilibrium (LD) $r^2 > 0.6$ in the 1000 Genomes EUR) in our ancestry-specific GWAS summary statistics. P-value thresholds were determined using the method of the effective degrees of freedom for the spectrally decomposed covariance matrix for the block of SNPs (53). Briefly, LD matrix were calculated for the block of SNPs with the appropriate ancestry from HaploReg database (54), specifically, EUR for European Americans, AFR for African Americans, AMR for Hispanic Americans and ASN for East-Asian Americans. Then, we estimated the effective degrees of freedom (N_{eff}) calculated from the spectrally decomposed covariance matrix for the block of SNPs, using the relationship $N_{eff} = \left(\sum_{k=1}^K \lambda_k\right)^2 / \left(\sum_{k=1}^K \lambda_k^2\right)$, where λ_k is the k^{th} eigen value of the $K \times K$ covariance matrix for the K SNPs (55). Finally, the nominal significance threshold (0.05) was divided by N_{eff} . If one SNP from the block of SNPs had a p-value smaller than the calculated threshold, the block was considered to be significantly replicated.

Fine mapping with haplotype blocks and 95% credible sets variants

We constructed the haplotype map of the genetic region that harbors the GWAS significant locus in each ancestry population. The haplotype block structure ± 5 kb from the GWAS lead SNP was determined based on haplotypes with $\pm 5\%$ occurrence, using the 4-gamete rule as implemented in Haploview 4.0 (56). The haplotype block sizes were compared among the ancestry groups.

To fine-map each locus showing genome-wide significance in the GWAS meta-analysis, we constructed the 95% credible sets which represent the minimum set of variants accounting for 95% of posterior probabilities in the region (57). Assuming a single causal variant for each locus and that the true causal variant is either genotyped or imputed, the probability that the 95% credible set contains the causal variant would be around 0.95. We first calculated the posterior probabilities for each variant located ± 500 kb from the top variant (variant showing the lowest P-value in the region), i.e. the corresponding Bayes Factor (BF) divided by the summation of BF over all variants in the region. Variants were then ranked according to their BF and the ranked variants were combined to form the credible set until their cumulative posterior probability attains or exceeds 95%.

Functional annotation of credible set variants

We annotated the credible set variants for regulatory features against the HaploReg database (54, 58). Specifically, we explored evidence of transcription factor binding, DNase hypersensitivity as well as promoter and enhancer activities via histone modification signals.

Active promoter is marked by signature of H3K4me3, whereas H3K27ac and H3K4me1 define regions with enhancer activity. Additionally, we conducted methylation quantitative loci (meQTL) analyses to assess whether the associated SNPs influences DNA methylation using datasets at birth, childhood, adolescence, pregnancy and middle age (59). The *cis*-meQTL SNPs from the HaploReg analysis were annotated using the Genotype-Tissue Expression (GTEx) database (60) to examine gene expression across several tissues.

Colocalization

To identify shared causal loci associated with lipid traits and gene expression, colocalization analysis was performed using the *coloc* R package (61). We used summary statistics from our *trans*-ancestry meta-analysis and expression quantitative trait loci (eQTL) based on the GTEx v.7 database in nine tissues relevant for lipid traits (liver, whole blood, artery aorta, artery coronary, adipose visceral omentum, heart left ventricle, adipose subcutaneous, heart atrial appendage, muscle skeletal) (60, 62). For each locus, the input data included summary statistics for all SNPs 250 kb on either side of each significant lead SNP from TransMeta. A posterior probability of association with both traits (lipid trait and eQTL genetic variant) higher than 75% was considered to be evidence of colocalization and was visualized using *LocusCompareR* R package (63).

RESULTS

Our analysis included 608 European American, 623 African American, 552 Hispanic American and 235 East Asian American pregnant women. The mean \pm s.d. maternal age was 30.4 ± 4.5 , 25.5 ± 5.3 , 27.0 ± 5.5 , and 30.7 ± 4.6 years, respectively (Table 1). Triglycerides levels differ by ancestry, with mean \pm s.d. of 120.4 ± 46.5 for European American, 105.2 ± 38.3 for African American, 141.1 ± 55.0 for Hispanic American and 142.2 ± 55.3 for East Asian American women (p -value = 4.9×10^{-11}), while other lipid concentrations were similar among ancestry groups.

Trans-ancestry meta-analysis

Trans-ancestry meta-analyses found three genome-wide significant associations: a locus in/near the *APOE-APOC1-TOMM40* gene (consisting 10 SNPs, with lead SNP rs7412, p -value = 6.86×10^{-17}) associated with total cholesterol; a locus near *CELSR2* gene (consisting 5 SNPs, with lead SNP rs7528419, p -value = 2.79×10^{-13}) and a locus near the *APOE-APOC1-TOMM40* gene (consisting 14 SNPs, with lead SNP rs7412, p -value = 9.83×10^{-30}) associated with LDL (Table 2, Figure 1, Supplementary Figure S2). No locus showed genome-wide significant association with HDL and triglycerides, but two SNPs associated with HDL in the well-known lipid loci (*ABCA1*, rs4149307, p -value = 9.71×10^{-8} and *CETP*, rs11076175, p -value = 2.97×10^{-7}) narrowly missed the significance threshold (Supplementary Table S1).

The haplotype blocks that harbor each of the genome-wide significant loci are presented in Supplementary Figures S4 and S6, and show that for the *CELSR2* gene locus (associated with LDL), the size of the haplotype block is smaller in African American than European American women (Supplementary Figure S6). In the 95% credible set analysis of the three

loci associated with maternal lipid traits, we were able to narrow down the putative causal SNPs down to the lead SNPs for total cholesterol and LDL (Supplementary Table S2). The SNPs in the credible set overlap with genomic regions showing enhancer activities of the histone modification signatures H3k27ac and H3K4me1 in tissues including liver, blood and placenta (Supplementary Table S3). Based on the mQTL database of methylation quantitative trait loci (meQTL) at serial time points across the life course (Supplementary Table S4), rs7528419 (*CELSR2*) was *cis*-meQTL in blood with the CpG methylation marker cg21684021 known to be associated with birth weight (64) and rs7412 was *cis*-meQTL with cg05644480 associated with gestational age (65). By querying the Genotype-Tissue Expression (GTEx) database we found that genes *APOE* and *APOC1* annotating SNPs in the 95% credible set are highly expressed in liver (Supplementary Figure S7).

Evaluation of published lipids loci in pregnant women

In our trans-ancestry meta-analysis, 100 out of 148 (68.9%) previously known total cholesterol-related loci, 26 out of 44 (59.1%) LDL-related loci, 121 out of 165 (73.3%) HDL-related loci, 100 out of 145 (69.0%) triglycerides-related loci had consistent direction, and 29 (19.6%), 1 (2.3%), 32 (19.4%), and 26 (17.9%) loci were replicated in pregnant women (consistent direction and p-value < 0.05 in our data; Figure 2 and Supplementary Table S5). Local replication identified additional replicated SNPs with an improvement across ancestries (Figure 3 and Supplementary Table S6).

Variance explained

We calculated the variance explained by our significant lead SNPs and previously published related-lipid loci. The variance explained by our lead SNPs ranged from 2.7% for total cholesterol among Hispanic Americans to 12.8% for LDL among European Americans, with small differences in variances observed across ancestry by lipid traits (Table 3). The previously published lipid loci explained from 0% (total cholesterol among East Asian Americans) to 33.6% (HDL among Hispanic Americans) of the variance in the lipid traits. Specifically total cholesterol and triglycerides had higher variance explained among European Americans, while LDL and HDL had higher variance explained among East Asian Americans.

Colocalization

We found strong evidence of colocalization between LDL in early pregnancy and *CELSR2* gene expression in liver and skeletal muscle (Posterior probabilities (PP) of 90% of a shared causal variant) (Supplementary Table S7). The best predicted causal variant was the rs7528419 GWAS lead SNP replicated by our analysis or another SNP in strong LD with the SNP (rs12740374, $r^2=1$ in European population and 0.88 in African population) (Figure 4). Visualization of the 500kb region of rs7528419 [*CELSR2*] based on LDL summary statistics and GTEx gene expression eQTL in liver (Supplementary Figure S8) and skeletal muscle (Supplementary Figure S9) shows a pattern consistent with colocalization.

DISCUSSION

In the first *trans*-ancestry genomic study of lipid traits in pregnant women, we identified loci in two genes (*CELSR2* and *APOE*) reaching genome-wide significant association with early gestation levels of total cholesterol and LDL levels, and two other loci (*CETP* and *ABCA1*) approaching genome-wide significant association with HDL. We found that only 15% of lipid trait loci identified in predominantly European adults were transferable to pregnant women from diverse ancestries, with some improvement when implementing local replication approaches, underlining the need for genome-wide lipids-related association studies in pregnant women from diverse ancestries. Finally, colocalization analysis identified *CELSR2* as a candidate causal gene in which gene expression in liver and skeletal muscle and LDL blood level are attributable to a single causal genetic variant.

The four loci identified in our study (*APOE*, *ABCA1*, *CETP* and *CELSR2*) are well-known lipid trait-related genes (23, 24, 28, 66–73). Candidate gene studies have found associations of SNPs in *ABCA1* and *CETP* with plasma HDL levels during second and third trimesters in Chinese pregnant women (74, 75). *APOE* (Apolipoprotein E) is involved in lipoprotein-mediated lipid transport (76), and has been implicated in the onset and development of coronary artery atherosclerosis. *APOE* interacts with *ABCA1* (ATP-Binding cassette 1) to mediate cholesterol efflux pump in the cellular lipid removal pathway (77). Mutations in *ABCA1* cause Tangier disease and familial HDL deficiency (78, 79). *CETP* (Cholesteryl Ester Transfer Protein) is involved in the transfer of cholesteryl ester from HDL to other lipoproteins and regulates cholesterol transport (80), and *CETP* gene defect has been associated with hyperalphalipoproteinemia 1 characterized by high HDL levels (81).

Our colocalization analysis highlighted a potential causal SNP that may explain LDL blood levels and expression of *CELSR2* in liver and skeletal muscle. This is in line with recent GWAS study conducted among Hispanic populations that reported a causal SNP associated with LDL and total cholesterol in liver and skeletal muscle (73). Furthermore, *CELSR2* in liver has been previously identified as a candidate gene associated with coronary artery disease and plasma LDL levels (82, 83). *CELSR2* gene expression has also been associated with fat accumulation in liver tissue of patients with nonalcoholic fatty liver disease and in mice models (84), highlighting the tissue-specific *CELSR2* function in the regulation of lipid metabolism.

The proportion of known lipid loci directly replicated by our study was lower than reports from previously published studies in adults (24, 28, 31). Local replication increased the replication rate among non-European groups. The lower replication rates in our study may be due the small sample size, unique genetic architecture of lipid traits in pregnant women, and the ancestral diversity of our cohort unlike previous replication efforts in predominantly male European populations. This highlights the need for large-scale GWAS of lipid traits in pregnant women from diverse ancestry groups to understand the genetic architecture of lipids. The variance in lipid levels explained by a genetic risk score derived using the two significant loci in the present cohort appeared to be better correlated with lipid traits among pregnant women compared to a genetic risk score derived using all previously known lipid loci in non-pregnant populations. The prediction accuracy of these polygenic scores is likely

to improve as the number of lipid loci relevant for pregnant women increases with future studies.

Our study has several limitations. The sample size is modest for a GWAS. However, the presence of multi-ancestry individuals was a unique strength that facilitated trans-ancestry meta-analysis and identification of significant associations even with a modest sample size. Furthermore, using the colocalization analysis we were able to report a causal variant in *CELSR2* associated with LDL and gene expression in liver, confirming recent studies. However, future larger studies are needed to discover variants with low frequency and ancestry-specific effects. We did not detect loci significantly associated with early gestation levels of triglycerides potentially because triglyceride levels during pregnancy can fluctuate due to hormonal action (85). Moreover, compared to other lipids, triglycerides in women have been shown to be influenced by higher environmental factors; hence, the need for much larger samples to identify genetic effects (42). Another limitation was that lipid measures were limited to the first trimester, while lipids are known to rise during pregnancy (2). Future studies with longitudinal lipid sampling are needed to understand genetic regulation of lipid trajectories during pregnancy.

In conclusion, this trans-ancestry GWAS meta-analysis in pregnant women identified four known lipid-loci. Moreover, the results shed light on potential colocalization between LDL and *CELSR2* in liver in skeletal muscle. The low level of replication of known loci underlines the need for genome-wide lipid trait-related association studies in diverse populations.

Supplementary Material

Refer to Web version on PubMed Central for supplementary material.

Acknowledgments:

We acknowledge the study participants of the NICHD Fetal Growth Studies. We thank research teams at all participating clinical centers (which include Christina Care Health Systems, Columbia University, Fountain Valley Hospital, California, Long Beach Memorial Medical Center, New York Hospital, Queens, Northwestern University, University of Alabama at Birmingham, University of California, Irvine, Medical University of South Carolina, Saint Peters University Hospital, Tufts University, and Women and Infants Hospital of Rhode Island). The authors also acknowledge the Wadsworth Center, C-TASC and the EMMES Corporations in providing data and imaging support. This work utilized the computational resources of the NIH HPC Biowulf cluster (<http://hpc.nih.gov>).

Sources of Funding:

This research was supported by the Intramural Research Program of the *Eunice Kennedy Shriver* National Institute of Child Health and Human Development, National Institutes of Health including American Recovery and Reinvestment Act funding via contract numbers HHSN275200800013C; HHSN275200800002I; HHSN27500006; HHSN275200800003IC; HHSN275200800014C; HHSN275200800012C; HHSN275200800028C; HHSN275201000009C and HHSN27500008. Additional support was obtained from the NIH Office of the Director, the National Institute on Minority Health and Health Disparities, and the National Institute of Diabetes and Digestive and Kidney Diseases.

Data availability:

The maternal genotype data analyzed in the current study are available from the corresponding author upon request.

Abbreviations:

BF	Bayes Factor
eQTL	expression quantitative trait loci
GWAS	Genome-wide association studies
HDL	high-density lipoprotein cholesterol
LD	linkage disequilibrium
LDL	low-density lipoprotein cholesterol
meQTL	methylation quantitative loci
PCs	Principal components
SNPs	single nucleotide polymorphisms
s.d.	standard deviation

REFERENCES

1. Leiva A, Salsoso R, Saez T, Sanhueza C, Pardo F, Sobrevia L. Cross-sectional and longitudinal lipid determination studies in pregnant women reveal an association between increased maternal LDL cholesterol concentrations and reduced human umbilical vein relaxation. *Placenta*. 2015;36(8):895–902. Epub 2015/06/10. doi: 10.1016/j.placenta.2015.05.012. [PubMed: 26055528]
2. Grantz KL, Elmi A, Pugh SJ, Catov J, Sjaarda L, Albert PS. Maternal Serum Lipid Trajectories and Association with Pregnancy Loss and Length of Gestation. *Am J Perinatol*. 2019. Epub 2019/06/04. doi: 10.1055/s-0039-1689000.
3. Nasioudis D, Doulaveris G, Kanninen TT. Dyslipidemia in pregnancy and maternal-fetal outcome. *Minerva Ginecol*. 2019;71(2):155–62. Epub 2018/10/16. doi: 10.23736/S0026-4784.18.04330-7. [PubMed: 30318877]
4. El Khoully NI, Sanad ZF, Saleh SA, Shabana AA, Elhalaby AF, Badr EE. Value of first-trimester serum lipid profile in early prediction of preeclampsia and its severity: A prospective cohort study. *Hypertens Pregnancy*. 2016;35(1):73–81. Epub 2016/02/02. doi: 10.3109/10641955.2015.1115060. [PubMed: 26829675]
5. Bever AM, Mumford SL, Schisterman EF, Sjaarda L, Perkins NJ, Gerlanc N, et al. Maternal preconception lipid profile and gestational lipid changes in relation to birthweight outcomes. *Sci Rep*. 2020;10(1):1374. Epub 2020/01/30. doi: 10.1038/s41598-019-57373-z. [PubMed: 31992758]
6. Clausen T, Burski TK, Oyen N, Godang K, Bollerslev J, Henriksen T. Maternal anthropometric and metabolic factors in the first half of pregnancy and risk of neonatal macrosomia in term pregnancies. A prospective study. *Eur J Endocrinol*. 2005;153(6):887–94. Epub 2005/12/03. doi: 10.1530/eje.1.02034. [PubMed: 16322395]
7. Vrijkotte TG, Algra SJ, Brouwer IA, van Eijnsden M, Twickler MB. Maternal triglyceride levels during early pregnancy are associated with birth weight and postnatal growth. *J Pediatr*. 2011;159(5):736–42 e1. Epub 2011/06/28. doi: 10.1016/j.jpeds.2011.05.001. [PubMed: 21705016]
8. Wang X, Guan Q, Zhao J, Yang F, Yuan Z, Yin Y, et al. Association of maternal serum lipids at late gestation with the risk of neonatal macrosomia in women without diabetes mellitus. *Lipids Health Dis*. 2018;17(1):78. Epub 2018/04/13. doi: 10.1186/s12944-018-0707-7. [PubMed: 29642923]
9. Ouidir M, Mendola P, Workalemahu T, Grewal J, Grantz KL, Zhang C, et al. Race-ethnic differences in the associations of maternal lipid trait genetic risk scores with longitudinal fetal growth. *J Clin Lipidol*. 2019;13(5):821–31. Epub 2019/08/07. doi: 10.1016/j.jacl.2019.06.007. [PubMed: 31383602]

10. Napoli C, Palinski W. Maternal hypercholesterolemia during pregnancy influences the later development of atherosclerosis: clinical and pathogenic implications. *Eur Heart J*. 2001;22(1):4–9. Epub 2001/01/03. doi: 10.1053/euhj.2000.2147. [PubMed: 11133201]
11. Smedts HP, van Uitert EM, Valkenburg O, Laven JS, Eijkemans MJ, Lindemans J, et al. A derangement of the maternal lipid profile is associated with an elevated risk of congenital heart disease in the offspring. *Nutr Metab Cardiovasc Dis*. 2012;22(6):477–85. Epub 2010/12/28. doi: 10.1016/j.numecd.2010.07.016. [PubMed: 21186113]
12. Daraki V, Georgiou V, Papavasiliou S, Chalkiadaki G, Karahaliou M, Koinaki S, et al. Metabolic profile in early pregnancy is associated with offspring adiposity at 4 years of age: the Rhea pregnancy cohort Crete, Greece. *PLoS One*. 2015;10(5):e0126327. Epub 2015/05/15. doi: 10.1371/journal.pone.0126327. [PubMed: 25970502]
13. Napoli C, D'Armiento FP, Mancini FP, Postiglione A, Witztum JL, Palumbo G, et al. Fatty streak formation occurs in human fetal aortas and is greatly enhanced by maternal hypercholesterolemia. Intimal accumulation of low density lipoprotein and its oxidation precede monocyte recruitment into early atherosclerotic lesions. *J Clin Invest*. 1997;100(11):2680–90. Epub 1998/02/12. doi: 10.1172/JCI119813. [PubMed: 9389731]
14. Farias DR, Alves-Santos NH, Eshriqui I, Martins MC, Struchiner CJ, Lepsch J, et al. Leptin gene polymorphism (rs7799039; G2548A) is associated with changes in serum lipid concentrations during pregnancy: a prospective cohort study. *Eur J Nutr*. 2020;59(5):1999–2009. Epub 2019/07/12. doi: 10.1007/s00394-019-02049-7. [PubMed: 31292750]
15. Scifres CM, Catov JM, Simhan HN. The impact of maternal obesity and gestational weight gain on early and mid-pregnancy lipid profiles. *Obesity (Silver Spring)*. 2014;22(3):932–8. Epub 2013/07/16. doi: 10.1002/oby.20576. [PubMed: 23853155]
16. Schreuder YJ, Hutten BA, van Eijdsden M, Jansen EH, Vissers MN, Twickler MT, et al. Ethnic differences in maternal total cholesterol and triglyceride levels during pregnancy: the contribution of demographics, behavioural factors and clinical characteristics. *Eur J Clin Nutr*. 2011;65(5):580–9. Epub 2011/01/20. doi: 10.1038/ejcn.2010.282. [PubMed: 21245878]
17. van Dongen J, Willemsen G, Chen WM, de Geus EJ, Boomsma DI. Heritability of metabolic syndrome traits in a large population-based sample. *J Lipid Res*. 2013;54(10):2914–23. Epub 2013/08/07. doi: 10.1194/jlr.P041673. [PubMed: 23918046]
18. Weiss LA, Pan L, Abney M, Ober C. The sex-specific genetic architecture of quantitative traits in humans. *Nat Genet*. 2006;38(2):218–22. Epub 2006/01/24. doi: 10.1038/ng1726. [PubMed: 16429159]
19. Elder SJ, Lichtenstein AH, Pittas AG, Roberts SB, Fuss PJ, Greenberg AS, et al. Genetic and environmental influences on factors associated with cardiovascular disease and the metabolic syndrome. *J Lipid Res*. 2009;50(9):1917–26. Epub 2009/04/18. doi: 10.1194/jlr.P900033-JLR200. [PubMed: 19372593]
20. Pilia G, Chen WM, Scuteri A, Orru M, Albai G, Dei M, et al. Heritability of cardiovascular and personality traits in 6,148 Sardinians. *PLoS Genet*. 2006;2(8):e132. Epub 2006/08/29. doi: 10.1371/journal.pgen.0020132. [PubMed: 16934002]
21. Zarkesh M, Daneshpour MS, Faam B, Fallah MS, Hosseinzadeh N, Guity K, et al. Heritability of the metabolic syndrome and its components in the Tehran Lipid and Glucose Study (TLGS). *Genet Res (Camb)*. 2012;94(6):331–7. Epub 2013/02/05. doi: 10.1017/S001667231200050X. [PubMed: 23374242]
22. Kathiresan S, Manning AK, Demissie S, D'Agostino RB, Surti A, Guiducci C, et al. A genome-wide association study for blood lipid phenotypes in the Framingham Heart Study. *BMC Med Genet*. 2007;8 Suppl 1:S17. Epub 2007/10/16. doi: 10.1186/1471-2350-8-s1-s17. [PubMed: 17903299]
23. de Vries PS, Brown MR, Bentley AR, Sung YJ, Winkler TW, Ntalla I, et al. Multiancestry Genome-Wide Association Study of Lipid Levels Incorporating Gene-Alcohol Interactions. *Am J Epidemiol*. 2019;188(6):1033–54. Epub 2019/01/31. doi: 10.1093/aje/kwz005. [PubMed: 30698716]
24. Klarin D, Damrauer SM, Cho K, Sun YV, Teslovich TM, Honerlaw J, et al. Genetics of blood lipids among ~300,000 multi-ethnic participants of the Million Veteran Program. *Nat Genet*. 2018;50(11):1514–23. Epub 2018/10/03. doi: 10.1038/s41588-018-0222-9. [PubMed: 30275531]

25. Willer CJ, Schmidt EM, Sengupta S, Peloso GM, Gustafsson S, Kanoni S, et al. Discovery and refinement of loci associated with lipid levels. *Nat Genet.* 2013;45(11):1274–83. Epub 2013/10/08. doi: 10.1038/ng.2797. [PubMed: 24097068]
26. Liu DJ, Peloso GM, Yu H, Butterworth AS, Wang X, Mahajan A, et al. Exome-wide association study of plasma lipids in >300,000 individuals. *Nat Genet.* 2017;49(12):1758–66. Epub 2017/10/31. doi: 10.1038/ng.3977. [PubMed: 29083408]
27. Teslovich TM, Musunuru K, Smith AV, Edmondson AC, Stylianou IM, Koseki M, et al. Biological, clinical and population relevance of 95 loci for blood lipids. *Nature.* 2010;466(7307):707–13. Epub 2010/08/06. doi: 10.1038/nature09270. [PubMed: 20686565]
28. Below JE, Parra EJ, Gamazon ER, Torres J, Krithika S, Candille S, et al. Meta-analysis of lipid-traits in Hispanics identifies novel loci, population-specific effects, and tissue-specific enrichment of eQTLs. *Sci Rep.* 2016;6:19429. Epub 2016/01/20. doi: 10.1038/srep19429. [PubMed: 26780889]
29. Lu X, Peloso GM, Liu DJ, Wu Y, Zhang H, Zhou W, et al. Exome chip meta-analysis identifies novel loci and East Asian-specific coding variants that contribute to lipid levels and coronary artery disease. *Nat Genet.* 2017;49(12):1722–30. Epub 2017/10/31. doi: 10.1038/ng.3978. [PubMed: 29083407]
30. Albrechtsen A, Grarup N, Li Y, Sparso T, Tian G, Cao H, et al. Exome sequencing-driven discovery of coding polymorphisms associated with common metabolic phenotypes. *Diabetologia.* 2013;56(2):298–310. Epub 2012/11/20. doi: 10.1007/s00125-012-2756-1. [PubMed: 23160641]
31. Asselbergs FW, Guo Y, van Iperen EP, Sivapalaratnam S, Tragante V, Lanktree MB, et al. Large-scale gene-centric meta-analysis across 32 studies identifies multiple lipid loci. *Am J Hum Genet.* 2012;91(5):823–38. Epub 2012/10/16. doi: 10.1016/j.ajhg.2012.08.032. [PubMed: 23063622]
32. Chasman DI, Pare G, Mora S, Hopewell JC, Peloso G, Clarke R, et al. Forty-three loci associated with plasma lipoprotein size, concentration, and cholesterol content in genome-wide analysis. *PLoS Genet.* 2009;5(11):e1000730. Epub 2009/11/26. doi: 10.1371/journal.pgen.1000730. [PubMed: 19936222]
33. Peloso GM, Auer PL, Bis JC, Voorman A, Morrison AC, Stitzel NO, et al. Association of low-frequency and rare coding-sequence variants with blood lipids and coronary heart disease in 56,000 whites and blacks. *Am J Hum Genet.* 2014;94(2):223–32. Epub 2014/02/11. doi: 10.1016/j.ajhg.2014.01.009. [PubMed: 24507774]
34. Kuchenbaecker K, Telkar N, Reiker T, Walters RG, Lin K, Eriksson A, et al. The transferability of lipid loci across African, Asian and European cohorts. *Nat Commun.* 2019;10(1):4330. Epub 2019/09/26. doi: 10.1038/s41467-019-12026-7. [PubMed: 31551420]
35. Wierzbicki AS, Reynolds TM. Genetic risk scores in lipid disorders. *Curr Opin Cardiol.* 2019;34(4):406–12. Epub 2019/06/07. doi: 10.1097/HCO.0000000000000623. [PubMed: 31169601]
36. Tam CHT, Lim CKP, Luk AOY, Ng ACW, Lee HM, Jiang G, et al. Development of genome-wide polygenic risk scores for lipid traits and clinical applications for dyslipidemia, subclinical atherosclerosis, and diabetes cardiovascular complications among East Asians. *Genome Med.* 2021;13(1):29. Epub 2021/02/21. doi: 10.1186/s13073-021-00831-z. [PubMed: 33608049]
37. Palmisano BT, Zhu L, Eckel RH, Stafford JM. Sex differences in lipid and lipoprotein metabolism. *Mol Metab.* 2018. Epub 2018/06/03. doi: 10.1016/j.molmet.2018.05.008.
38. Wang X, Magkos F, Mittendorfer B. Sex differences in lipid and lipoprotein metabolism: it's not just about sex hormones. *J Clin Endocrinol Metab.* 2011;96(4):885–93. Epub 2011/04/09. doi: 10.1210/jc.2010-2061. [PubMed: 21474685]
39. Silander K, Alanne M, Kristiansson K, Saarela O, Ripatti S, Auro K, et al. Gender differences in genetic risk profiles for cardiovascular disease. *PLoS One.* 2008;3(10):e3615. Epub 2008/11/01. doi: 10.1371/journal.pone.0003615. [PubMed: 18974842]
40. McCarthy JJ. Gene by sex interaction in the etiology of coronary heart disease and the preceding metabolic syndrome. *Nutr Metab Cardiovasc Dis.* 2007;17(2):153–61. Epub 2007/02/20. doi: 10.1016/j.numecd.2006.01.005. [PubMed: 17306735]

41. Flynn E, Tanigawa Y, Rodriguez F, Altman RB, Sinnott-Armstrong N, Rivas MA. Sex-specific genetic effects across biomarkers. *Eur J Hum Genet.* 2021;29(1):154–63. Epub 20200901. doi: 10.1038/s41431-020-00712-w. [PubMed: 32873964]
42. Wong MWK, Thalamuthu A, Braidy N, Mather KA, Liu Y, Ciobanu L, et al. Genetic and environmental determinants of variation in the plasma lipidome of older Australian twins. *eLife.* 2020;9:e58954. doi: 10.7554/eLife.58954. [PubMed: 32697195]
43. Grewal J, Grantz KL, Zhang C, Sciscione A, Wing DA, Grobman WA, et al. Cohort Profile: NICHD Fetal Growth Studies-Singletons and Twins. *Int J Epidemiol.* 2018;47(1):25–1. Epub 2017/10/13. doi: 10.1093/ije/dyx161. [PubMed: 29025016]
44. Shrestha D, Workalemahu T, Tekola-Ayele F. Maternal dyslipidemia during early pregnancy and epigenetic ageing of the placenta. *Epigenetics.* 2019;14(10):1030–9. Epub 2019/06/11. doi: 10.1080/15592294.2019.1629234. [PubMed: 31179827]
45. Ouidir M, Zeng X, Workalemahu T, Shrestha D, Grantz KL, Mendola P, et al. Early pregnancy dyslipidemia is associated with placental DNA methylation at loci relevant for cardiometabolic diseases. *Epigenomics.* 2020;12(11):921–34. Epub 2020/07/18. doi: 10.2217/epi-2019-0293. [PubMed: 32677467]
46. Friedewald WT, Levy RI, Fredrickson DS. Estimation of the concentration of low-density lipoprotein cholesterol in plasma, without use of the preparative ultracentrifuge. *Clin Chem.* 1972;18(6):499–502. Epub 1972/06/01. [PubMed: 4337382]
47. Tekola-Ayele F, Zhang C, Wu J, Grantz KL, Rahman ML, Shrestha D, et al. Trans-ethnic meta-analysis of genome-wide association studies identifies maternal ITPR1 as a novel locus influencing fetal growth during sensitive periods in pregnancy. *PLoS Genet.* 2020;16(5):e1008747. Epub 2020/05/15. doi: 10.1371/journal.pgen.1008747. [PubMed: 32407400]
48. Li Y, Willer CJ, Ding J, Scheet P, Abecasis GR. MaCH: using sequence and genotype data to estimate haplotypes and unobserved genotypes. *Genet Epidemiol.* 2010;34(8):816–34. Epub 2010/11/09. doi: 10.1002/gepi.20533. [PubMed: 21058334]
49. Shi J, Lee S. A novel random effect model for GWAS meta-analysis and its application to trans-ethnic meta-analysis. *Biometrics.* 2016;72(3):945–54. Epub 2016/02/27. doi: 10.1111/biom.12481. [PubMed: 26916671]
50. Pruim RJ, Welch RP, Sanna S, Teslovich TM, Chines PS, Gliedt TP, et al. LocusZoom: regional visualization of genome-wide association scan results. *Bioinformatics.* 2010;26(18):2336–7. Epub 2010/07/17. doi: 10.1093/bioinformatics/btq419. [PubMed: 20634204]
51. Tange O GNU Parallel 20200622 ('Floyd'). Zenodo. 2020:zenodo.3903853. doi: 10.5281/zenodo.3903853.
52. Magi R, Morris AP. GWAMA: software for genome-wide association meta-analysis. *BMC Bioinformatics.* 2010;11:288. Epub 2010/06/01. doi: 10.1186/1471-2105-11-288. [PubMed: 20509871]
53. Adeyemo AA, Tekola-Ayele F, Doumatey AP, Bentley AR, Chen G, Huang H, et al. Evaluation of Genome Wide Association Study Associated Type 2 Diabetes Susceptibility Loci in Sub Saharan Africans. *Front Genet.* 2015;6:335. Epub 2015/12/05. doi: 10.3389/fgene.2015.00335. [PubMed: 26635871]
54. Ward LD, Kellis M. HaploReg: a resource for exploring chromatin states, conservation, and regulatory motif alterations within sets of genetically linked variants. *Nucleic Acids Res.* 2012;40(Database issue):D930–4. Epub 2011/11/09. doi: 10.1093/nar/gkr917. [PubMed: 22064851]
55. Ramos E, Chen G, Shriner D, Doumatey A, Gerry NP, Herbert A, et al. Replication of genome-wide association studies (GWAS) loci for fasting plasma glucose in African-Americans. *Diabetologia.* 2011;54(4):783–8. Epub 2010/12/29. doi: 10.1007/s00125-010-2002-7. [PubMed: 21188353]
56. Barrett JC, Fry B, Maller J, Daly MJ. Haploview: analysis and visualization of LD and haplotype maps. *Bioinformatics.* 2005;21(2):263–5. Epub 2004/08/07. doi: 10.1093/bioinformatics/bth457. [PubMed: 15297300]

57. Wellcome Trust Case Control C, Maller JB, McVean G, Byrnes J, Vukcevic D, Palin K, et al. Bayesian refinement of association signals for 14 loci in 3 common diseases. *Nat Genet.* 2012;44(12):1294–301. Epub 2012/10/30. doi: 10.1038/ng.2435. [PubMed: 23104008]
58. Ward LD, Kellis M. HaploReg v4: systematic mining of putative causal variants, cell types, regulators and target genes for human complex traits and disease. *Nucleic Acids Res.* 2016;44(D1):D877–81. Epub 2015/12/15. doi: 10.1093/nar/gkv1340. [PubMed: 26657631]
59. Gaunt TR, Shihab HA, Hemani G, Min JL, Woodward G, Lyttleton O, et al. Systematic identification of genetic influences on methylation across the human life course. *Genome Biol.* 2016;17:61. Epub 2016/04/03. doi: 10.1186/s13059-016-0926-z. [PubMed: 27036880]
60. Consortium G. The Genotype-Tissue Expression (GTEx) pilot analysis: multitissue gene regulation in humans. *Science.* 2015;348(6235):648–60. [PubMed: 25954001]
61. Giambartolomei C, Vukcevic D, Schadt EE, Franke L, Hingorani AD, Wallace C, et al. Bayesian test for colocalisation between pairs of genetic association studies using summary statistics. *PLoS Genet.* 2014;10(5):e1004383. Epub 2014/05/17. doi: 10.1371/journal.pgen.1004383. [PubMed: 24830394]
62. Consortium G. The GTEx Consortium atlas of genetic regulatory effects across human tissues. *Science.* 2020;369(6509):1318–30. Epub 2020/09/12. doi: 10.1126/science.aaz1776. [PubMed: 32913098]
63. Liu B, Gloude-mans MJ, Rao AS, Ingelsson E, Montgomery SB. Abundant associations with gene expression complicate GWAS follow-up. *Nat Genet.* 2019;51(5):768–9. Epub 2019/05/03. doi: 10.1038/s41588-019-0404-0. [PubMed: 31043754]
64. Kupers LK, Monnereau C, Sharp GC, Yousefi P, Salas LA, Ghantous A, et al. Meta-analysis of epigenome-wide association studies in neonates reveals widespread differential DNA methylation associated with birthweight. *Nat Commun.* 2019;10(1):1893. Epub 2019/04/25. doi: 10.1038/s41467-019-09671-3. [PubMed: 31015461]
65. Hannon E, Schendel D, Ladd-Acosta C, Grove J, Hansen CS, Hougaard DM, et al. Variable DNA methylation in neonates mediates the association between prenatal smoking and birth weight. *Philos Trans R Soc Lond B Biol Sci.* 2019;374(1770):20180120. Epub 2019/04/11. doi: 10.1098/rstb.2018.0120.
66. Proust C, Empana JP, Boutouyrie P, Alivon M, Challande P, Danchin N, et al. Contribution of Rare and Common Genetic Variants to Plasma Lipid Levels and Carotid Stiffness and Geometry: A Substudy of the Paris Prospective Study 3. *Circ Cardiovasc Genet.* 2015;8(4):628–36. Epub 2015/07/15. doi: 10.1161/CIRCGENETICS.114.000979. [PubMed: 26160806]
67. Tang CS, Zhang H, Cheung CY, Xu M, Ho JC, Zhou W, et al. Exome-wide association analysis reveals novel coding sequence variants associated with lipid traits in Chinese. *Nat Commun.* 2015;6:10206. Epub 2015/12/23. doi: 10.1038/ncomms10206. [PubMed: 26690388]
68. Kim YJ, Go MJ, Hu C, Hong CB, Kim YK, Lee JY, et al. Large-scale genome-wide association studies in East Asians identify new genetic loci influencing metabolic traits. *Nat Genet.* 2011;43(10):990–5. Epub 2011/09/13. doi: 10.1038/ng.939. [PubMed: 21909109]
69. Rasmussen-Torvik LJ, Pacheco JA, Wilke RA, Thompson WK, Ritchie MD, Kho AN, et al. High density GWAS for LDL cholesterol in African Americans using electronic medical records reveals a strong protective variant in APOE. *Clin Transl Sci.* 2012;5(5):394–9. Epub 2012/10/17. doi: 10.1111/j.1752-8062.2012.00446.x. [PubMed: 23067351]
70. Tabassum R, Ramo JT, Ripatti P, Koskela JT, Kurki M, Karjalainen J, et al. Genetic architecture of human plasma lipidome and its link to cardiovascular disease. *Nat Commun.* 2019;10(1):4329. Epub 2019/09/26. doi: 10.1038/s41467-019-11954-8. [PubMed: 31551469]
71. Andaleon A, Mogil LS, Wheeler HE. Gene-based association study for lipid traits in diverse cohorts implicates BACE1 and SIDT2 regulation in triglyceride levels. *PeerJ.* 2018;6:e4314. Epub 2018/02/07. doi: 10.7717/peerj.4314. [PubMed: 29404214]
72. Oni-Orisan A, Haldar T, Ranatunga DK, Medina MW, Schaefer C, Krauss RM, et al. The impact of adjusting for baseline in pharmacogenomic genome-wide association studies of quantitative change. *NPJ Genom Med.* 2020;5:1. Epub 2020/01/24. doi: 10.1038/s41525-019-0109-4. [PubMed: 31969989]

73. Andaleon A, Mogil LS, Wheeler HE. Genetically regulated gene expression underlies lipid traits in Hispanic cohorts. *PLoS One*. 2019;14(8):e0220827. Epub 2019/08/09. doi: 10.1371/journal.pone.0220827. [PubMed: 31393916]
74. Cui M, Li W, Ma L, Ping F, Liu J, Wu X, et al. HDL-cholesterol concentration in pregnant Chinese Han women of late second trimester associated with genetic variants in CETP, ABCA1, APOC3, and GALNT2. *Oncotarget*. 2017;8(34):56737–46. Epub 2017/09/17. doi: 10.18632/oncotarget.18128. [PubMed: 28915626]
75. Nie M, Wang Y, Li W, Ping F, Liu J, Wu X, et al. The association between six genetic variants and blood lipid levels in pregnant Chinese Han women. *J Clin Lipidol*. 2017;11(4):938–44. Epub 2017/07/02. doi: 10.1016/j.jacl.2017.06.006. [PubMed: 28666712]
76. Marcel YL, Vezina C, Milne RW. Cholesteryl ester and apolipoprotein E transfer between human high density lipoproteins and chylomicrons. *Biochim Biophys Acta*. 1983;750(2):411–7. Epub 1983/02/07. doi: 10.1016/0005-2760(83)90047-4. [PubMed: 6860692]
77. Krimbou L, Denis M, Haidar B, Carrier M, Marcil M, Genest J Jr. Molecular interactions between apoE and ABCA1: impact on apoE lipidation. *J Lipid Res*. 2004;45(5):839–48. Epub 2004/02/03. doi: 10.1194/jlr.M300418-JLR200. [PubMed: 14754908]
78. Brooks-Wilson A, Marcil M, Clee SM, Zhang LH, Roomp K, van Dam M, et al. Mutations in ABC1 in Tangier disease and familial high-density lipoprotein deficiency. *Nat Genet*. 1999;22(4):336–45. Epub 1999/08/04. doi: 10.1038/11905. [PubMed: 10431236]
79. Marcil M, Brooks-Wilson A, Clee SM, Roomp K, Zhang LH, Yu L, et al. Mutations in the ABC1 gene in familial HDL deficiency with defective cholesterol efflux. *Lancet*. 1999;354(9187):1341–6. Epub 1999/10/26. doi: 10.1016/s0140-6736(99)07026-9. [PubMed: 10533863]
80. Qiu X, Mistry A, Ammirati MJ, Chrnyk BA, Clark RW, Cong Y, et al. Crystal structure of cholesteryl ester transfer protein reveals a long tunnel and four bound lipid molecules. *Nat Struct Mol Biol*. 2007;14(2):106–13. Epub 2007/01/24. doi: 10.1038/nsmb1197. [PubMed: 17237796]
81. Brown ML, Inazu A, Hesler CB, Agellon LB, Mann C, Whitlock ME, et al. Molecular basis of lipid transfer protein deficiency in a family with increased high-density lipoproteins. *Nature*. 1989;342(6248):448–51. Epub 1989/11/23. doi: 10.1038/342448a0. [PubMed: 2586614]
82. Schadt EE, Molony C, Chudin E, Hao K, Yang X, Lum PY, et al. Mapping the genetic architecture of gene expression in human liver. *PLoS Biol*. 2008;6(5):e107. Epub 2008/05/09. doi: 10.1371/journal.pbio.0060107. [PubMed: 18462017]
83. Kathiresan S, Melander O, Guiducci C, Surti A, Burtt NP, Rieder MJ, et al. Six new loci associated with blood low-density lipoprotein cholesterol, high-density lipoprotein cholesterol or triglycerides in humans. *Nat Genet*. 2008;40(2):189–97. Epub 2008/01/15. doi: 10.1038/ng.75. [PubMed: 18193044]
84. Tan J, Che Y, Liu Y, Hu J, Wang W, Hu L, et al. CELSR2 deficiency suppresses lipid accumulation in hepatocyte by impairing the UPR and elevating ROS level. *FASEB J*. 2021;35(10):e21908. Epub 2021/09/04. doi: 10.1096/fj.202100786RR. [PubMed: 34478580]
85. Ghio A, Bertolotto A, Resi V, Volpe L, Di Cianni G. Chapter 7 - Triglyceride metabolism in pregnancy. In: Makowski GS, editor. *Advances in Clinical Chemistry*. 55: Elsevier; 2011. p. 133–53. [PubMed: 22126027]

Highlights

- This first *trans*-ancestry GWAS of lipids in pregnant women identified 4 known loci.
- Loci in *CELSR2* and *APOE* were associated with levels of total cholesterol and LDL.
- Loci in *CETP* and *ABCA1* approached the genome-wide significance with HDL levels.
- Local replication analysis underlined the need for studies in diverse populations.
- Colocalization analysis identified *CELSR2* as a candidate causal gene.

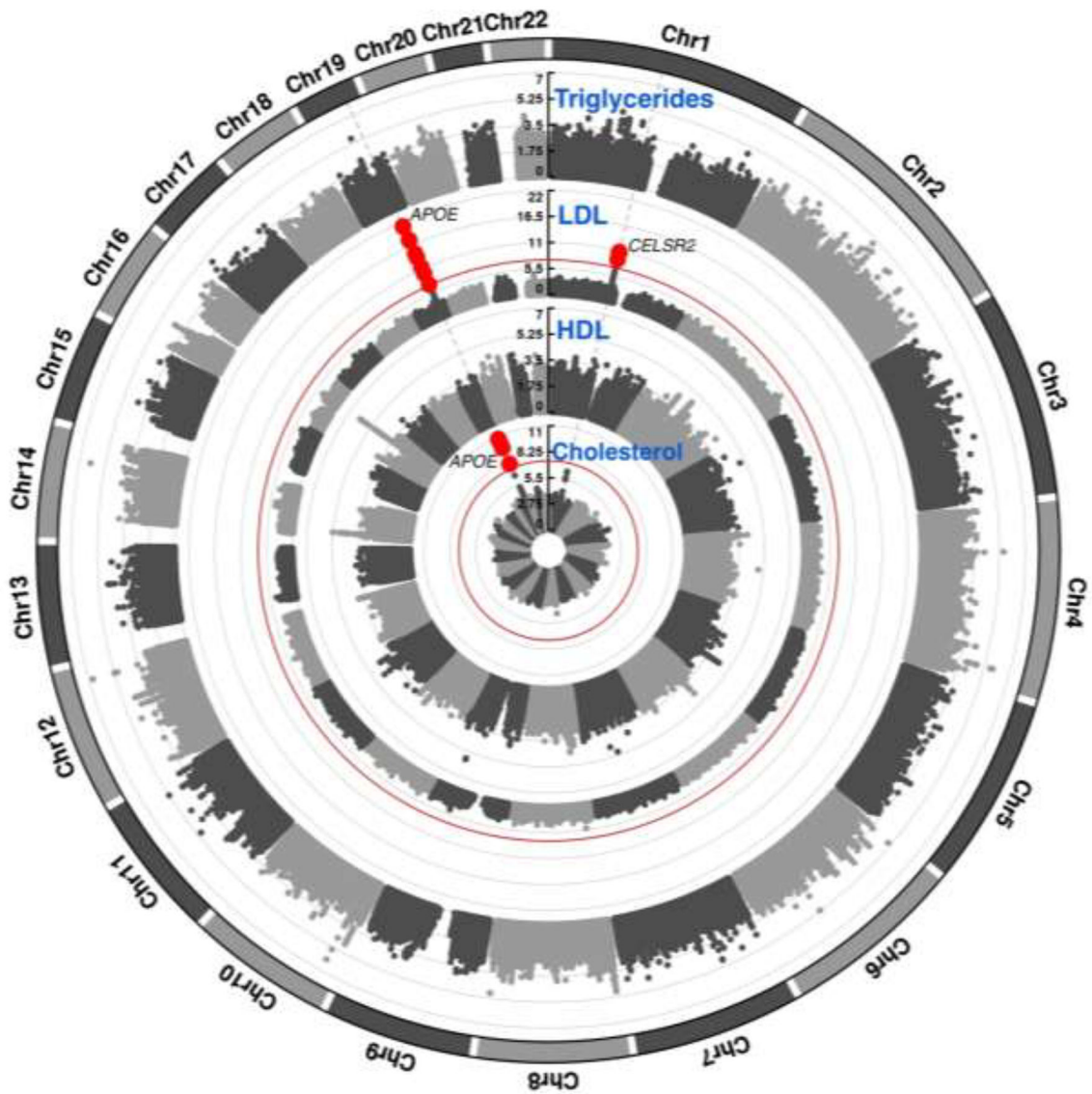


Figure 1: Circular Manhattan plot of genome-wide trans-ancestry meta-analyses associations. Log₁₀ p-values are plotted across 22 chromosomes for five lipid traits. SNPs surpassing genome-wide significant association ($p\text{-value} < 5 \times 10^{-8}$) are marked in red and nearby genes are included.

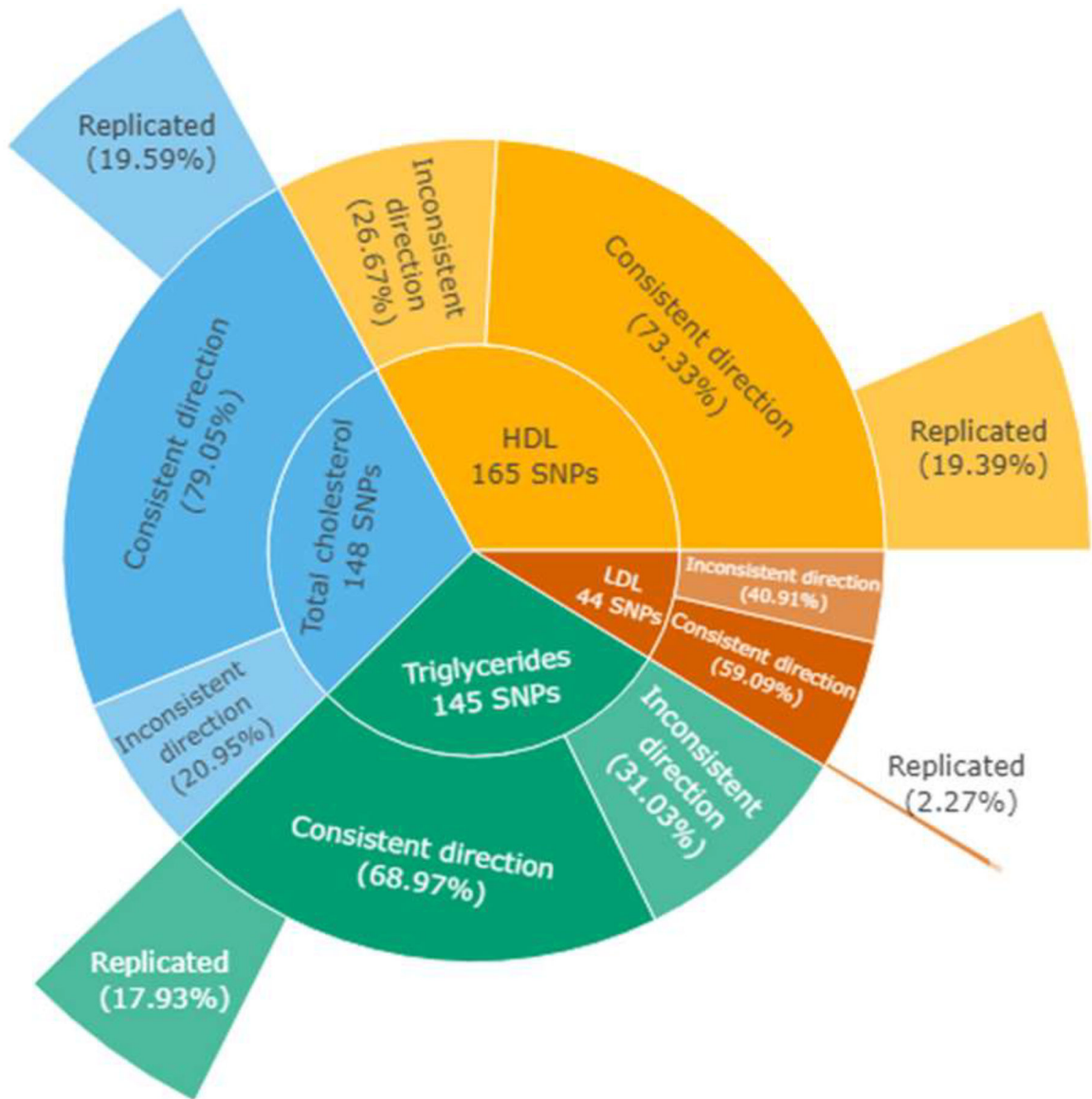


Figure 2: Transferability of previously known lipid loci in pregnant women. The inner-most pie shows the number of previously known loci associated with each of the four lipid traits. The middle pie shows the percentage of SNPs with directionally consistent effect in our data. The outermost pie shows the percentage of replicated SNPs (with both directionally consistent effect and p-values < 0.05 in our data).

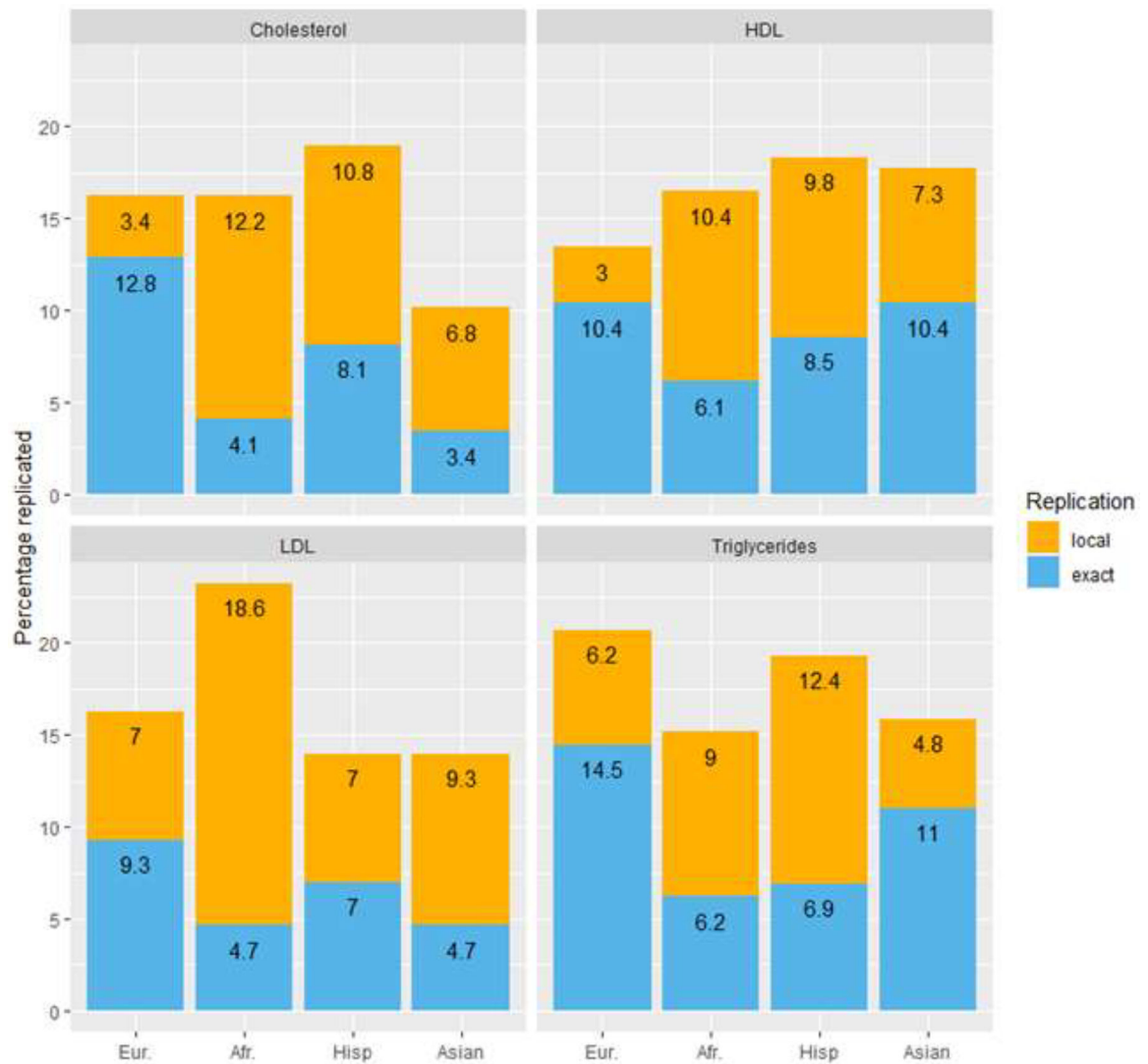


Figure 3: Ancestry-specific local replication of previously known lipid trait-related SNPs. Local replication was assessed in our GWAS summary statistics for all SNPs within linkage disequilibrium ($LD > 0.6$) from the known index SNP.

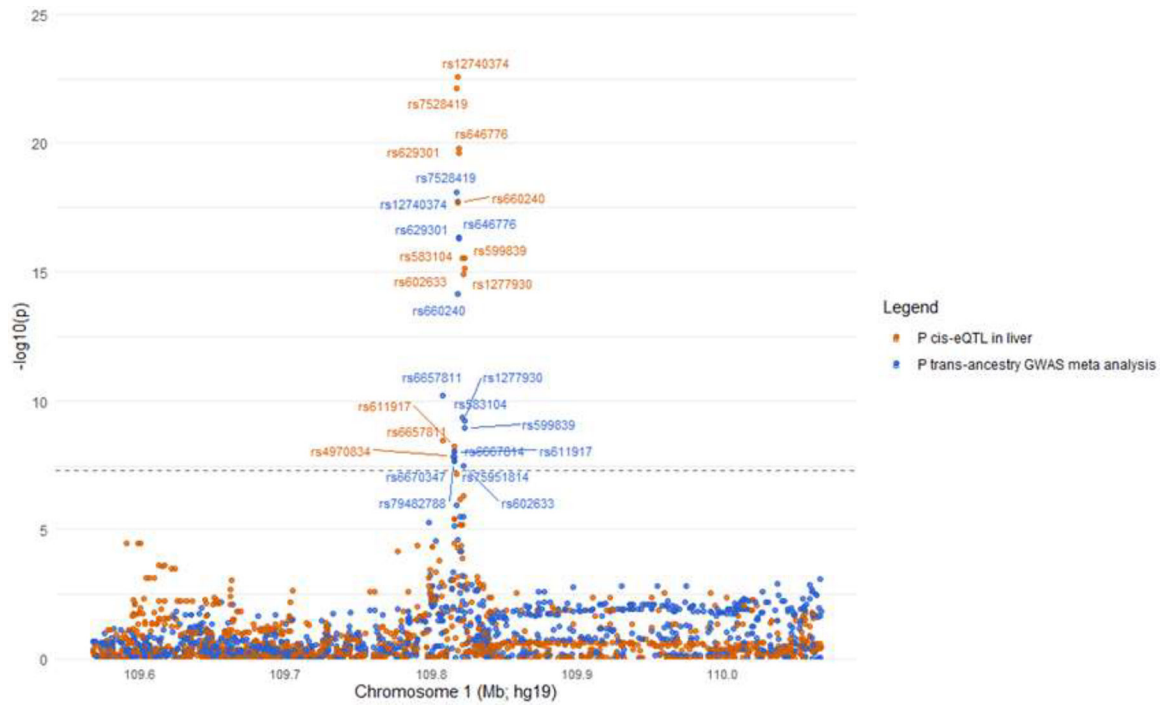


Figure 4: *CELSR2* locus zoom plot of trans-ancestry meta-analysis associations for low density lipoprotein cholesterol (LDL) and cis-eQTL associations with *CELSR2* gene expression in liver.

Table 1:

Description of pregnant women from the Fetal Growth Studies - Singleton included in the study (N= 1654).

	European American n = 608		African American n = 623		Hispanic American n = 552		East Asian American n = 235		P-value*
	mean	sd	mean	sd	mean	sd	mean	sd	
Maternal characteristics									
Maternal age, years	30.4	4.5	25.5	5.3	27.0	5.5	30.7	4.6	1.4×10^{-74}
Pre-pregnancy BMI, kg/m ²	25.0	5.1	26.7	5.7	26.0	4.9	22.1	2.7	6.6×10^{-33}
Gestational age at enrollment, weeks	12.5	1.0	12.5	1.1	12.9	0.9	12.9	0.8	1.7×10^{-17}
Cholesterol, mg/dL	189.8	31.0	179.9	31.3	185.2	31.2	182.8	28.1	0.06
HDL, mg/dL	63.4	13.4	62.0	14.4	58.0	13.7	63.0	12.6	0.07
LDL**, mg/dL	102.3	27.3	97.0	27.0	99.1	25.5	91.4	24.0	0.96
Triglycerides, mg/dL	120.4	46.5	105.2	38.3	141.1	55.0	142.2	55.3	4.9×10^{-11}

* P-values derived from analysis of variance or χ^2 test comparing 4 groups.

** 3 women with missing LDL values (1 European American, 1 African American and 1 Hispanic American)

Abbreviations: HDL: High-density lipoprotein cholesterol, LDL: Low-density lipoprotein cholesterol.

Table 2:

Genome-wide significant associations (p-value < 5*10⁻⁸) from the trans-ancestry meta-analysis of lipid traits in pregnant women.

Lipid Traits	SNP ID	chr: position hg19	TransMeta				GWAMA							
			Optimal rho	P-value	Effect allele	EAF	beta	[95% CI]	se	p.value	n studies	n samples	effects*	Referen allele
Total cholesterol	rs1160983	19:45397229	0.09	1.70E-09	G	0.945	15.42	[11.17, 19.66]	2.17	1.18E-12	4	2018	++++	G
	rs61679753	19:45400747	0.25	2.69E-13	T	0.936	17.44	[13.49, 21.39]	2.01	5.10E-18	4	2018	++++	T
	rs111784051	19:45402262	0.25	2.90E-13	T	0.933	17.16	[13.29, 21.04]	1.98	4.28E-18	4	2018	++++	T
	rs429358	19:45411941	0.25	7.86E-13	T	0.841	-11.66	[-14.31, -9.01]	1.35	7.03E-18	4	2018	----	T
	rs7412	19:45412079	0.09	6.86E-17	C	0.927	17.89	[14.33, 21.45]	1.82	7.72E-23	4	2018	++++	C
	rs1065853	19:45413233	0.09	1.12E-16	G	0.927	17.69	[14.14, 21.25]	1.81	1.65E-22	4	2018	++++	G
	rs72654473	19:45414399	0.25	8.39E-11	C	0.895	12.23	[9.16, 15.31]	1.57	6.93E-15	4	2018	++++	C
	rs814573	19:45424351	0.09	3.28E-08	A	0.707	-6.72	[-9, -4.44]	1.16	7.81E-09	4	2018	----	T
	rs190712692	19:45425178	0.09	9.66E-16	G	0.942	19.29	[15.28, 23.3]	2.05	4.49E-21	4	2018	++++	G
rs141622900	19:45426792	0.09	1.15E-15	G	0.942	19.18	[15.18, 23.18]	2.04	5.85E-21	4	2018	++++	G	
Calculated LDL	rs7528419	1:109817192	0.09	2.79E-13	A	0.777	8.49	[6.55, 10.43]	0.99	9.72E-18	4	2015	++++	A
	rs12740374	1:109817590	0.09	5.09E-13	G	0.783	8.55	[6.59, 10.51]	1.00	1.22E-17	4	2015	++++	G
	rs660240	1:109817838	0.09	1.42E-10	T	0.250	-7.37	[-9.27, -5.47]	0.97	3.17E-14	4	2015	----	T
	rs629301	1:109818306	0.09	4.92E-12	G	0.259	-7.76	[-9.63, -5.89]	0.95	4.22E-16	4	2015	----	G
	rs646776	1:109818530	0.09	4.62E-12	C	0.258	-7.84	[-9.72, -5.96]	0.96	3.23E-16	4	2015	----	C
	rs7254892	19:45389596	1	1.30E-10	G	0.933	12.88	[9.65, 16.12]	1.65	6.50E-15	4	2015	++++	G
	rs1160983	19:45397229	0.09	4.11E-15	G	0.945	17.16	[13.5, 20.82]	1.87	3.91E-20	4	2015	++++	G
	rs61679753	19:45400747	0.09	1.31E-20	T	0.936	19.07	[15.67, 22.46]	1.73	4.11E-28	4	2015	++++	T
	rs111784051	19:45402262	0.25	7.93E-21	T	0.933	18.92	[15.58, 22.26]	1.70	1.11E-28	4	2015	++++	T
	rs769446	19:45408628	1	2.61E-09	T	0.932	13.04	[9.5, 16.57]	1.80	5.20E-13	4	2015	++++	T
	rs769449	19:45410002	1	2.39E-08	G	0.930	-11.02	[-14.22, -7.83]	1.63	1.31E-11	4	2015	----	G
	rs429358	19:45411941	0.25	1.50E-18	T	0.841	-12.28	[-14.56, -10]	1.16	4.66E-26	4	2015	----	T
	rs7412	19:45412079	0.09	9.83E-30	C	0.927	20.86	[17.79, 23.94]	1.57	2.53E-40	4	2015	++++	C

Author Manuscript

Author Manuscript

Author Manuscript

Author Manuscript

Lipid Traits	SNP ID	chr: position hg19	TransMeta				GWAMA									Referen allele
			Optimal rho	P-value	Effect allele	EAf	beta	[95% CI]	se	p-value	n studies	n samples	effects*			
	rs1065853	19:45413233	0.09	1.68E-29	G	0.927	20.67	[17.6, 23.73]	1.56	8.28E-40	4	2015	++++	G		
	rs72654473	19:45414399	0.09	1.08E-16	C	0.895	13.46	[10.8, 16.12]	1.36	3.57E-23	4	2015	++++	C		
	rs12721051	19:45422160	0.09	2.63E-09	C	0.883	-9.14	[-11.7, -6.58]	1.31	2.58E-12	4	2015	----	C		
	rs814573	19:45424351	1	1.45E-09	A	0.707	-7.32	[-9.27, -5.36]	1.00	2.21E-13	4	2015	----	A		
	rs190712692	19:45425178	0.09	2.70E-25	G	0.942	21.28	[17.83, 24.74]	1.76	1.46E-33	4	2015	++++	G		
	rs141622900	19:45426792	0.09	4.08E-25	G	0.942	21.13	[17.69, 24.58]	1.76	2.70E-33	4	2015	++++	G		

Nearest gene	GWAS															
	European American				African American				Hispanic American				East Asian American			
	EAf	beta	se	p-values	EAf	beta	se	p-values	EAf	beta	se	p-values	EAf	beta	se	p-values
	0.96	24.03	4.81	5.76E-07	0.91	11.83	3.15	1.75E-04	0.97	19.07	5.52	5.51E-04	0.94	11.78	5.26	2.51E-02
	0.96	23.90	4.77	5.43E-07	0.89	15.92	2.93	5.40E-08	0.96	18.87	4.98	1.54E-04	0.92	13.84	4.69	3.15E-03
	0.96	23.86	4.77	5.56E-07	0.88	15.40	2.83	5.25E-08	0.96	18.98	4.97	1.35E-04	0.92	13.96	4.65	2.66E-03
	0.86	-11.13	2.59	1.66E-05	0.77	-12.59	2.15	4.92E-09	0.87	-9.81	2.84	5.39E-04	0.89	-13.51	4.18	1.24E-03
	0.92	19.16	3.19	1.95E-09	0.91	18.22	3.03	1.80E-09	0.96	18.05	4.46	5.25E-05	0.92	14.18	4.69	2.48E-03
<i>APOE</i> (583 bases downstream)	0.92	19.14	3.19	2.06E-09	0.90	18.00	2.99	1.76E-09	0.96	17.43	4.50	1.08E-04	0.92	14.12	4.69	2.60E-03
<i>APOE</i> (1749 bases downstream)	0.90	14.86	2.89	2.84E-07	0.85	12.02	2.43	7.20E-07	0.94	7.60	3.75	4.30E-02	0.92	13.38	4.70	4.42E-03
	0.82	-11.21	2.47	5.50E-06	0.55	-6.20	1.87	9.10E-04	0.71	-3.27	2.15	1.29E-01	0.82	-8.94	3.74	1.68E-02
<i>APOC1</i> (2572 bases downstream)	0.94	22.81	3.88	3.94E-09	0.93	20.29	3.43	3.33E-09	0.97	17.16	5.22	1.01E-03	0.91	14.51	4.45	1.10E-03
<i>APOC1P1</i> (3268 bases upstream)	0.94	22.76	3.88	4.35E-09	0.93	20.34	3.44	3.19E-09	0.97	17.16	5.21	9.97E-04	0.91	14.14	4.38	1.25E-03
	0.76	6.86	1.79	1.29E-04	0.72	7.75	1.66	3.05E-06	0.78	11.73	1.83	1.61E-10	0.94	4.82	4.47	2.81E-01
	0.76	6.86	1.79	1.29E-04	0.74	8.15	1.70	1.58E-06	0.79	11.35	1.84	7.01E-10	0.94	5.18	4.53	2.53E-01
	0.23	-6.64	1.84	2.94E-04	0.36	-6.91	1.57	1.09E-05	0.23	-9.01	1.79	5.04E-07	0.06	-5.18	4.53	2.53E-01
	0.24	-6.80	1.79	1.48E-04	0.37	-7.14	1.58	5.98E-06	0.24	-9.93	1.74	1.04E-08	0.06	-4.61	4.32	2.86E-01
	0.24	-6.80	1.79	1.49E-04	0.37	-7.26	1.59	4.72E-06	0.24	-9.92	1.74	1.30E-08	0.06	-5.18	4.53	2.53E-01
	0.96	22.67	4.04	2.05E-08	0.88	9.70	2.30	2.36E-05	0.96	10.91	3.93	5.54E-03	0.94	15.47	4.43	4.83E-04
	0.96	25.30	4.24	2.45E-09	0.91	12.02	2.74	1.19E-05	0.97	21.06	4.53	3.26E-06	0.94	17.98	4.48	5.95E-05
	0.96	25.10	4.21	2.46E-09	0.89	16.56	2.55	8.24E-11	0.96	20.01	4.09	9.81E-07	0.92	18.87	3.99	2.25E-06
	0.96	25.06	4.21	2.53E-09	0.88	16.32	2.46	3.46E-11	0.96	20.15	4.08	7.85E-07	0.92	19.03	3.96	1.51E-06
	0.90	10.62	2.76	1.17E-04	0.96	16.38	4.25	1.17E-04	0.95	10.84	4.04	7.29E-03	0.91	17.56	4.11	1.97E-05
	0.89	-12.98	2.47	1.40E-07	0.97	-8.51	4.97	8.65E-02	0.94	-7.83	3.18	1.37E-02	0.91	-12.37	3.70	8.29E-04
	0.86	-12.02	2.28	1.38E-07	0.77	-12.66	1.88	1.44E-11	0.87	-10.64	2.33	5.02E-06	0.89	-15.40	3.56	1.51E-05

Author Manuscript

Author Manuscript

Author Manuscript

Author Manuscript

Nearest gene	GWAS															
	European American				African American				Hispanic American				East Asian American			
	EAF	beta	se	p-values	EAF	beta	se	p-values	EAF	beta	se	p-values	EAF	beta	se	p-values
	0.92	22.67	2.83	1.08E-15	0.91	20.00	2.64	3.42E-14	0.96	20.95	3.66	1.04E-08	0.92	19.16	3.99	1.55E-06
<i>APOE</i> (583 bases downstream)	0.92	22.66	2.83	1.13E-15	0.90	19.46	2.60	7.77E-14	0.96	21.01	3.69	1.27E-08	0.92	19.13	3.99	1.63E-06
<i>APOE</i> (1749 bases downstream)	0.90	17.20	2.56	1.84E-11	0.85	11.63	2.11	3.63E-08	0.94	8.80	3.10	4.48E-03	0.92	18.66	4.00	3.09E-06
	0.82	-11.17	2.08	8.13E-08	0.91	-5.04	2.71	6.24E-02	0.92	-8.52	2.83	2.58E-03	0.87	-10.95	3.25	7.58E-04
	0.82	-11.62	2.18	9.17E-08	0.55	-6.27	1.63	1.14E-04	0.71	-4.36	1.77	1.39E-02	0.82	-11.62	3.18	2.58E-04
<i>APOC1</i> (2572 bases downstream)	0.94	25.47	3.42	9.38E-14	0.93	20.79	2.99	3.33E-12	0.97	21.58	4.28	4.55E-07	0.91	16.71	3.78	1.01E-05
<i>APOC1P1</i> (3268 bases upstream)	0.94	25.40	3.42	1.11E-13	0.93	20.81	2.99	3.43E-12	0.97	21.61	4.28	4.34E-07	0.91	16.21	3.73	1.38E-05

* effect order is: European American, African American, Hispanic American, East Asian American

Lead SNPs are highlighted in bold

LDL: Low-density lipoprotein cholesterol

EAF: effect allele frequency

se: standard error

Author Manuscript

Author Manuscript

Author Manuscript

Author Manuscript

Table 3:

Variance in lipid traits during pregnancy explained by lead SNPs from our analysis and previously published lipid trait-related loci.

lipid	SNPs tested	European American n = 608	African American n = 623	Hispanic American n = 552	East Asian American n = 235
Total cholesterol	Lead SNPs: Chr19:45412079 (rs7412)	0.059	0.054	0.027	0.038
LDL	Lead SNPs: Chr1:109817192 (rs7528419) and Chr19:45412079 (rs7412)	0.128	0.122	0.127	0.102
HDL	NA	NA	NA	NA	NA
Triglycerides	NA	NA	NA	NA	NA
Total cholesterol	list of 148 known total cholesterol-related loci	0.142	0.014	0.140	-0.001
LDL	list of 44 known LDL-related loci	0.033	0.019	0.067	0.069
HDL	list of 166 known HDL-related loci	0.084	0.047	0.109	0.336
Triglycerides	list of 147 known triglycerides-related loci	0.092	0.088	0.026	0.076

NA not available

LDL: Low-density lipoprotein cholesterol

HDL: High-density lipoprotein cholesterol